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SPECIFICATION FOR TRYPTONE, MICROBIOLOGICAL GRADE

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SPECIFICATION FOR TRYPTONE, MICROBIOLOGICAL GRADE

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SPECIFICATION FOR TRYPTONE, MICROBIOLOGICAL GRADE

0. FOREWORD

0.1 This Indian Standard was adopted by the Indian Standards Institution on 26 November 1973, after the draft finalized by the Food Hygiene, Sampling and Analysis Sectional Committee had been approved by the Agricultural and Food Products Division Council.

0.2 Unless the ingredients used in media for microbiological work are of uniform quality, the results obtained might be erroneous and might be unreliable. Since the media used in different laboratories often differ greatly in their quality, the results of microbiological work at different laboratories cannot be compared. Therefore, with a view to unifying the practices of different laboratories dealing with microbiology and providing guidance to the indigenous manufacturers regarding the quality, it was decided to bring out a series of Indian Standard specifications for ingredients commonly used in media for microbiological work.

0.3 Tryptone is a tryptic hydrolysate of casein. It may be used in any formulation where pancreas or trypsin digested casein is specified. It is an extremely rich source of amino-acid nitrogen and has a high tryptophan content.

0.4 For the purpose of deciding whether a particular requirement of this standard is complied with, the final value, observed or calculated, expressing the result of a test, shall be rounded off in accordance with IS: 2-1960*. The number of significant places retained in the rounded off value should be the same as that of the specified value in this standard.

1. SCOPE

1.1 This standard prescribes requirements and methods of test for tryptone, microbiological grade.

2. REQUIREMENTS

2.1 Tryptone, microbiological grade, shall completely dissolve in water.

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*Rules for rounding off numerical values (*revised*).

2.2 One percent solution in distilled water after autoclaving at 121°C for 30 minutes shall have *p*H between 5·0 and 7·0.

2.3 One percent aqueous solution having *p*H adjusted to 7·0 and after filtration and sterilization by autoclaving at 121°C for 30 minutes shall be clear.

2.4 It shall be able to support and shall not inhibit the growth of micro-organisms when it is incorporated in suitable medium as the sole growth supporting substance. The general guidelines for testing this characteristic are given in **19** of IS : 6854-1973*. With these general guidelines, micro-organisms like *Staphylococcus aureus* or *Escherichia coli* may be used as the test micro-organisms.

2.5 It shall not contain any viable micro-organisms when sterilized at 121°C for 30 minutes and tested according to the method given in IS : 5402-1969†.

2.6 It shall give a positive indole reaction when tested in accordance with the method prescribed in Appendix A.

2.7 The material shall also conform to the requirements given in Table 1.

3. PACKING, STORAGE AND MARKING

3.1 Packing — The material shall be securely packed in well filled wide-mouth containers with tight-fitting lids.

3.2 Storage — The material shall be stored in a cool and dry place.

3.3 Marking — Each container shall be marked legibly to give the following information:

a) Name of the material including the words 'Microbiological Grade',
b) Name and address of the manufacturer,
c) Minimum net content, and
d) Batch or code number.

3.3.1 The container may also be marked with the ISI Certification Mark.

NOTE — The use of the ISI Certification Mark is governed by the provisions of the Indian Standards Institution (Certification Marks) Act and the Rules and Regulations made thereunder. The ISI Mark on products covered by an Indian Standard conveys the assurance that they have been produced to comply with the requirements of that standard under a well-defined system of inspection, testing and quality control which is devised and supervised by ISI and operated by the producer. ISI marked products are also continuously checked by ISI for conformity to that standard as a further safeguard. Details of conditions under which a licence for the use of the ISI Certification Mark may be granted to manufacturers or processors, may be obtained from the Indian Standards Institution.

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*Methods of sampling and test for ingredients used in media for microbiological work.

†Method for plate count of bacteria in foodstuffs.

TABLE 1 REQUIREMENTS FOR TRYPTONE, MICROBIOLOGICAL GRADE

(*Clause* 2.7)

SL No.	CHARACTERISTIC	REQUIREMENT	METHOD OF TEST, REF TO	
			Cl No. of IS : 6854-1973*	Appendix of IS : 7128-1973†
(1)	(2)	(3)	(4)	(5)
i)	Moisture, percent by mass, *Max*	7	4	—
ii)	Ash, percent by mass, *Max*	15	6	—
iii)	Amino-acid nitrogen, percent by mass, *Min*	3	8	—
iv)	Total nitrogen, percent by mass, *Min*	13	9	—
v)	Phosphorus (as P), percent by mass, *Max*	1·3	—	E
vi)	Sodium chloride, percent by mass, *Max*	7·0	11	—
vii)	Iron (as Fe), mg/kg, *Max*	75	13	—
viii)	Copper (as Cu), mg/kg, *Max*	10	15	—
ix)	Zinc (as Zn), mg/kg, *Max*	25	18	—
x)	Fermentable carbohydrates	Nil	20	—

*Methods of sampling and test for ingredients used in media for microbiological work.

†Specification for proteose peptone, microbiological grade.

4. SAMPLING

4.1 The representative samples of the material shall be drawn according to the method prescribed in **3** of IS : 6854-1973*.

5. TESTS

5.1 Tests shall be carried out by the methods prescribed in **2** and in col 4 and 5 of Table 1.

5.2 Quality of Reagents — Unless specified otherwise, pure chemicals and distilled water (*see* IS : 1070-1960†) shall be employed in tests.

NOTE — 'Pure chemicals' shall mean chemicals that do not contain impurities which affect the results of analysis.

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*Methods of sampling and test for ingredients used in media for microbiological work.

†Specification for water, distilled quality (*revised*).

APPENDIX A

(*Clause* 2.6)

METHOD OF TEST FOR INDOLE REACTION

A-1. REAGENTS

A-1.1 Peptone Water Medium — Dissolve by gentle heating 10·0 g of peptone under test and 5·0 g of sodium chloride in 1 000 ml of distilled water. Adjust *p*H between 8·0 and 8·4 and boil for 10 minutes. Cool and filter through filter paper and adjust the *p*H between 7·2 and 7·4. Place required quantities in tubes and sterilize at 120°C for 15 minutes.

A-1.2 Kavac's Reagent — Dissolve 5·0 g of *p*-dimethylaminobenzaldehyde in 75 ml of amyl or isoamyl alcohol by heating at 50 to 55°C on water bath. Cool and add slowly 25·0 ml of concentrated hydrochloric acid (sp gr 1·18, *Min*). Keep the reagent either in an amber coloured bottle or in a bottle wrapped with black paper to protect it from light. When not in use, keep at 4°C in a refrigerator. The colour of the reagent should be yellow to light brown.

A-2. PROCEDURE

A-2.1 Inoculate a tube of peptone water medium with a bacterial strain known to produce indole from peptone. A suitable strain is *Escherichia coli*. Inoculate another tube of peptone water medium with a bacterial strain known not to produce indole from peptone. Suitable strains are *Enterobacter Cloacae* and *Salmonella typhi*. Incubate the inoculated tubes at 37°C for 48 hours. Add to the growth 0·5 ml of Kovac's reagent, shake well and examine after 1 minute. Red colour in the reagent layer indicates the presence of indole.